A SURVEY FOR *SPATHULA TRYSSA* BALL AND OTHER FRESHWATER FLATWORMS IN THE VICTORIAN ALPS WITH AN EVALUATION OF THE CONSERVATION STATUS OF EACH SPECIES

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Eight species of flatworm were identified during a survey of 7 alpine areas in 1993, including 3 species of *Spathula* (Tricladida : Dugesiidae), 4 undescribed species and *Reynoldsonia reynoldsoni* Ball (Tricladida : Dugesiidae). *Spathula tryssa* Ball was relatively common and widespread in two adjacent areas, despite being listed under the *Flora and Fauna Guarantee Act* 1988. Current levels of land use do not appear to be threatening the species with extinction, although cattle grazing may be a localised problem. The IUCN conservation status of the species should be seen as Lower Risk Least Concern. Of the remaining species, two (*S. gourbaultae* Ball and an undescribed species) have very restricted distributions on Mt Donna Buang and Mt Stirling respectively, and should be considered to be Vulnerable and Critically Endangered respectively. Three of the species (*S. agelaea* Hall & Ball, *R. reynoldsoni* and a further undescribed species) appear to be either widespread or secure, deserving a Lower Risk Least Concern category, one undescribed species has insufficient data to determine a conservation status (Data Deficient) and the final species (undescribed) requires further taxonomic investigation before conclusions can be drawn.

THE genus *Spathula* (Tricladida : Dugesiidae) consists of nine species restricted to high altitude areas of Australasia (Victoria, New South Wales and New Zealand). All species appear to have a preference for cool, well oxygenated mountain waters (Ball 1977). Of the 6 species of *Spathula* that occur in Victoria, all but one have apparently very restricted distributions, with *S. gourbaultae* Ball found only on Mt Donna Buang, *S. truculenta* Ball from Mt McKay (but also from Mt Kosciusko), *S. foeni* Ball from a single site at Buxton and *S. camara* Ball from Falls Creek. The remaining species, *S. tryssa* Ball, has only been found at two spring localities on Mt Buller (Ball 1977; Hay & Ball 1979). One record near Mt Howitt (Powling & Sedgley 1984) was identified only as *Spathula* cf. *tryssa*, but was made without sectioning and the specimen has been lost (D. Hay, pers. comm.). The final species, *S. agelaea* Hay & Ball, appears to be more widespread, found on Mt Buffalo and the Falls Creek–Mt Hotham area (Hay & Ball 1979).

S. tryssa is a medium size flatworm (around 8 mm in length) and differs from most other species of the genus by the lack of pigment (a characteristic shared only with *S. gourbaultae*), giving it a white appearance in the field and the lack of eyes (shared only in Victoria with the pigmented *S. camara*). These characteristics do not allow formal identification in the field, but external characters can be used to identify specimens in the laboratory. Formal identification of *Spathula* species is only possible by serial sectioning of properly preserved specimens.

On the basis of the limited distribution and perceived threats from alpine resort development and grazing, *S. tryssa* was listed under Schedule 2 of the *Flora and Fauna Guarantee Act* 1988. In 1993, a comprehensive survey for *S. tryssa* was conducted to better determine the distribution and conservation status of the species and to collect information on the preferred habitat and ecology of *S. tryssa* and other flatworm species that may occur with it. This paper presents the results of that survey.

SURVEY METHODS

Seventy survey sites for freshwater flatworms were identified from maps and local knowledge (a list of sites is available from the senior author). Sites were concentrated in 7 main areas: Mt Buller–Mt Stirling; Mt Hotham; Mt Howitt; Mt Buffalo; Lake Mountain; Mt Donna Buang; and Mt Baw Baw (Fig. 1). Sampling sites were more concentrated in the Mt Buller–Mt Stirling area, the major area where *S. tryssa* was known to occur.

Sites were sampled in December 1993. Emphasis was placed on collecting white unpigmented species to maximise the inclusion of *S. tryssa*. Emphasis was also placed on springs as this was the habitat of previously collected specimens (Hay & Ball 1979; Powling & Sedgley 1984), however other habitats were also searched to see if they were utilised. At each site, rocks and other in-stream habitat were searched by eye. Rocks were removed from the stream and the underside also searched. Specimens of flatworms remaining on hard sub-

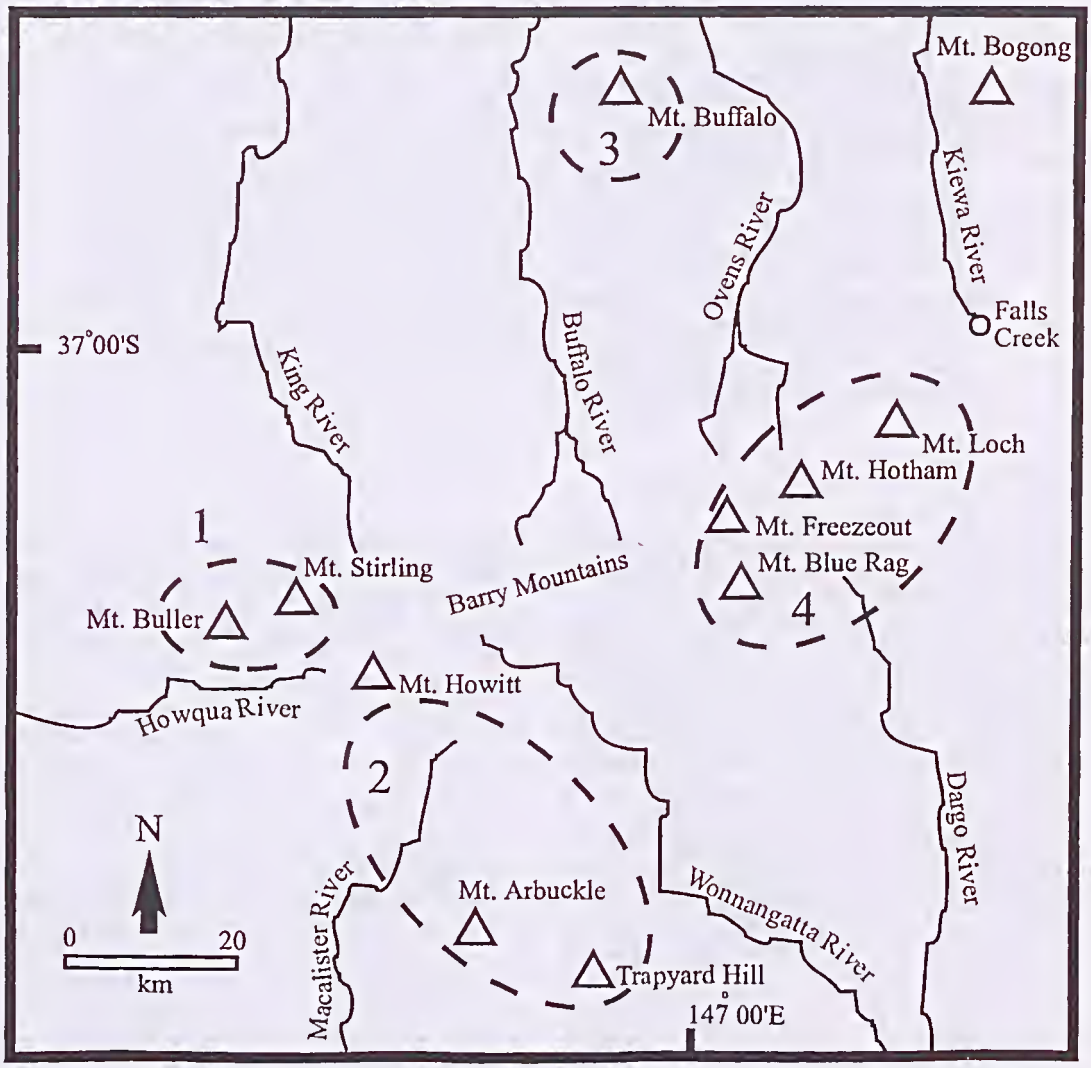

Fig. 1. Map of the central alps of Victoria showing the location of the 4 main sampling areas. More isolated areas to the south west (Mt Baw Baw, Mt Donna Buang and Lake Mountain) are not shown.

strate after removal from the water were covered by fixative solution, so they relaxed in a relatively flat position, and then sluiced off or removed using a fine brush. Animals on soft substrate were collected by brush or fine pipette and placed straight into fixative. As it is not possible to identify species in the field, specimens were killed in Dawar's solution (Dawar 1973) as this preserves much of the external morphology very well, as well as providing good fixation of internal anatomy for sectioning. Specimens were kept in Dawar's solution for about 24 hours before being transferred to alcohol.

IDENTIFICATION OF SPECIMENS

All specimens collected were identified using external morphology. Features used were color, presence or absence of eyes and number and position of sensory pits and fossae. These were sometimes comparatively subtle characters but could be seen with care. Some of these characters have previously been considered to be only apparent in sectioned specimens (eg. Ball 1977) but the use of Dawar's solution enabled these characters to be seen, greatly improving the identification of specimens.

The identification of selected specimens was confirmed by sectioned material. Twenty-one specimens of *S. tryssa*, 7 specimens of Species 1 and 1 specimen each of *S. gourbaultae*, *S. agelaea* and Species 2 were sectioned. Whole specimens were preserved in alcohol and were lodged with a few specimens on slides in the Museum of Victoria, Melbourne.

RESULTS

Species found during this study

Eight species of flatworm were collected and identified from 58 of the 70 sites surveyed (Table 1). This included three species of *Spathula* (*S. tryssa*, *S. gourbaultae* and *S. agelaea*). Another unpigmented dugesiid species *Reynoldsonia reynoldsoni* Ball was also identified, and significantly, four previously undescribed species (designated Species 1–4: Table 2) were located. Although some of the undescribed species appear to be similar to existing *Spathula* species, we have not ascribed a generic designation to them. Another large white flatworm belonging to a different order (Alloeocoelia, family Prorynchidae) was also found, but is not discussed in detail here.

Distribution of flatworm species

Spathula tryssa

S. tryssa was found at 17 of the 26 sites sampled around the Mt Buller–Mt Stirling area and at 7 of the 10 sites in the adjacent Howitt–Wellington massif (areas 1 and 2 in Fig. 1). On both these highland areas, the species could be found relatively easily in small spring-fed water bodies above 1475 m, often in high abundances. Examination of sectioned specimens from this study indicated that specimens in both these alpine areas were the same species. The two known localities on Mt Buller recorded in the literature could not be identified exactly, due to the large number of sites where the flatworms were present and the relatively vague nature of the original site descriptions (eg. the type

Collection area	Number of sites	*try*	*age*	*gou*	*Rey*	*1*	*2*	*3*	*4*
Mt Buller	17	12	0	0	0	0	0	0	0
Mt Stirling	5	3	0	0	0	2	0	0	0
Corn Hill	4	2	0	0	0	0	0	0	0
Howitt–Wellington area	10	7	0	0	0	0	0	2	0
Mt Baw Baw	1	0	0	0	0	0	0	0	1
Mt Donna Buang	1	0	0	1	0	0	0	0	0
Mt Hotham–Dinner Plain	15	0	1	0	0	0	4	3	9
Dargo High Plains	10	0	1	0	0	0	5	4	1
Mt Buffalo	5	0	1	0	5	0	0	0	0
Lake Mountain	1	0	0	0	0	0	0	0	1
Mt Skene	1	0	0	0	0	0	0	0	0
Total	70	24	3	1	5	2	9	9	11

Table 1. Distribution of flatworms collected during this study. *try* = *S. tryssa*, *age* = *S. agelaea*, *gou* = *S. gourbaultae*, *Rey* = *R. reynoldsoni*, *1* = Species 1, *2* = Species 2, *3* = Species 3, *4* = Species 4.

locality is described by Ball (1977) as 'Headwaters of Chalet Creek at 4600 ft on Mt Buller'). No specimens of *S. tryssa* were identified from the 34 sites sampled in other alpine areas, suggesting that the species is restricted to these two contiguous areas.

Spathula agelaea

S. agelaea was present at 3 sites, widely spread on Mt Buffalo, Mt Hotham–Dinner Plain and Dargo High Plains (areas 3 and 4 in Fig. 1), confirming the distribution noted by Hay & Ball (1979). This species was described by Hay & Ball (1979) from asexual specimens as they were unable to find any that were sexually mature. One sectioned specimen of *S. agelaea* was sexually mature and confirmed its specific uniqueness. At the time they suggested the species may be an asexual race of *S. tryssa*, but the sectioned specimen was clearly not *S. tryssa*.

Spathula gourbaultae

S. gourbaultae was confined to Mt Donna Buang, found only at the one locality sampled on that mountain.

Reynoldsonia reynoldsoni

R. reynoldsoni was found only from 5 sites, all on Mt Buffalo (area 3 in Fig. 1).

Species 1

The undescribed Species 1 was found only at two adjacent springs about 50 m apart on Mt Stirling (area 1 in Fig. 1). At one site it was found with *S. tryssa*. As it was not recorded at any of the other 26 sites in the Mt Buller–Mt Stirling area, it would appear to be the species with the most restricted distribution found in this study.

Species 2

Species 2 was moderately common around Mt Hotham–Dinner Plain and the Dargo High Plains (area 4 in Fig. 1), being found at 9 of the 25 sites with records spread over the entire sampling area.

Species 3

Species 3 occurred at 9 of the 35 sampling sites in both the Mt Howitt and Mt Hotham area (areas 2 and 4 in Fig. 1).

Species 4

Species 4 was found at 11 widespread sites, most commonly in the Mt Hotham–Dinner Plains area, but also on the Baw Baw Plateau, Dargo High Plains and Lake Mountain. As this species is probably a complex of species, taxonomic problems need to be resolved before distributional data can be further examined.

DISCUSSION

Distribution, species associations, habitat and ecology of Spathula tryssa

The data collected here suggest that *S. tryssa* is far more widespread than originally thought. While previously only recorded from the streams near the

Species	Description
Species 1	A pigmented species with an unpigmented ventral surface, and possessing obvious eyes. This species is very similar to *Spathula camara* in internal anatomy and sectioning of more specimens is required to differentiate the two. However, the presence of eyes distinguished it from all the pigmented species, including *S. camara*.
Species 2	An unpigmented species similar to *S. tryssa* but differing from it in, among other things, possessing only one pair of sensory pits and testes confined to the pre-pharyngeal region (*S. tryssa* has 3 pairs of pits and testes situated through the body length).
Species 3	A pigmented species. The combination of pigment, 2 pairs of ciliated pits, 1 pair of sensory fossae and no eyes suggest it to be a distinct, yet undescribed species.
Species 4	A pigmented species with a paler undersurface, varying from very pale to very dark grey. The variability in color suggest this designation may include more than one species, including *S. camara*.

Table 2. Description of undescribed Species 1–4 found during the study.

summit of Mt Buller, the species is now known to exist over a large area of the Howitt–Wellington massif (area 2 in Fig. 1). It also suggests that the lost specimen identified as *Spathula* cf. *tryssa* (Powling & Sedgley 1984) may well have been of that species (although due to the discovery of several undescribed species in this survey, this cannot be confirmed).

S. tryssa was found predominantly in spring-fed waters. Where these waters emerge from the ground, they are very cool and very shallow, often no more than a damp seep. The flatworms were easiest to collect in the alpine herbfield and in forest where the spring or seep had collected into pools or runnels, either naturally or in roadside gutters. *S. tryssa* was also found, but generally in low or moderate numbers, in creeks although they had possibly washed in from the spring-fed habitats further upstream. No specimens of *S. tryssa* were found in spring or creek habitats below about 1470 m, despite the apparent similarity of many of the lower altitude sites.

S. tryssa was usually the only flatworm species in the spring, but on occasions occurred with the prorynchid and at one locality with Species 1 and two localities with Species 3. With both Species 1 and Species 3, individuals of the two species were found clumped together under a single rock.

The species was abundant at several of the localities listed. At the site where it was most abundant, Macalister Springs, hundreds of individuals were found under one rock and flatworms were found under many rocks, as well as wandering on the substrate and water surface. The total population at this site was extremely large. The specimens were also much larger at this site, approximately twice the size of individuals at any other site.

The conclusion by Hay & Ball (1979) that the species retreats into the groundwater to escape drying of the surface water would appear to be valid. Most water bodies where specimens were collected would clearly dry up in most years, maybe several times, and flatworms are unable to withstand desiccation. Drying of potential habitat reduced the number of areas that could be sampled during this study on the south side of Mt Buller, Mt Stirling and parts of the Howitt High Plains area. Use of the groundwater would also explain the widespread occurrence of the species in the Buller–Corn Hill–Stirling area as flatworms could easily move into all available water bodies in the groundwater. Movement between such small, short-lived water bodies would otherwise be very difficult for a freshwater flatworm.

Hay & Ball (1979) state *S. tryssa* never experiences temperatures greater than 9°C and their laboratory study found viability of the species at 10°C to be very low. This lead them to suggest that *S. tryssa* retreated into the groundwater to avoid drying and high temperatures. During the current study specimens were found in higher temperatures, up to 19°C, although these high temperature habitats may have been areas that were about to dry out and had been disconnected from the groundwater, stranding the flatworms. As such, the temperature may not be representative of the habitat of successful flatworms.

The restriction to altitudes over 1470 m, if influencing distribution below ground as well as above ground, means that the population in the Mt Howitt–Mt Wellington area has been genetically distinct from the Mt Buller area populations for at least 15 thousand years and possibly up to 20 million years. Even within each of the two broad areas it is possible that populations are isolated and unable to interbreed, so that the population on Mt Stirling may now be genetically distinct from those of Corn Hill and Mt Buller. Populations in the Mt Howitt area may be even more disjointed.

Surface waters were often degraded after leaving the spring by collecting in roadside gutters, on ski runs or due to grazing by cattle. While found in roadside gutters and on ski runs, the flatworms were rarely found in areas where grazing had occurred. On Mt Stirling, no flatworms were found in the most trampled spring seep area. Both species of flatworms on Mt Stirling were found in areas that were amongst slightly higher vegetation which may suggest they need cover for protection or cooling. Grazing and wandering horses may reduce this cover and may be responsible for the absence of the flatworms in the trampled area but the evidence is not conclusive. In the Mt Howitt area, the absence of flatworms from trampled springs may also indicate a loss of habitat due to grazing.

Conservation status of Spathula tryssa

S. tryssa is restricted to two main adjacent areas of alpine Victoria. Taxonomic work suggests it is the same species in both main areas although only a few specimens have been sectioned in this and previous studies.

Based on the IUCN categories (IUCN 1994), there are no quantitative measures of population reduction, abundance or probability of extinction for *S. tryssa*, but the extent of occurrence is relatively large (perhaps extending over 2000 km^2).

Hence the category of Critically Endangered is not appropriate, but Endangered (extent of occurrence less than 5000 km^2) or Vulnerable (less than 20 000 km^2) may be appropriate.

The movement of the species through the groundwater means that the number of populations or subpopulations cannot be estimated, but is likely to be large. While local extinction seems to have occurred in several individual sites, possibly due to grazing, this cannot be seen as the same as extinction of a location or population as only part of the groundwater pool would have been disturbed and recolonisation of disturbed habitats would be expected to be relatively easy. Hence while the quality of the habitat is degraded at several points, there is no evidence for a decline in the area of occupancy, a key requirement for the designation of a conservation status.

In conclusion, *S. tryssa* is common and widespread on two mountain massifs, although probably as distinct populations. Several populations occur within the Alpine National Park although they may be locally adversely affected by grazing within the park. *S. tryssa* is apparently unaffected by other current land use practices. The greatest threats locally are damage to areas where the groundwater reaches the surface and over a larger scale, alteration to the groundwater quality and quantity. It seems that currently the species is not threatened with extinction and should be placed in the Lower Risk Least Concern category. As a result of the data collected during this survey, the species has been delisted under the *Flora and Fauna Guarantee Act* 1988.

Conservation status of other species

Spathula gourbaultae

This species is confined to Mt Donna Buang. Although listed as common by Hay & Ball (1979), specimens were found at only one locality during this study. This may be due to the absence of groundwater at the time or may indicate reduction in numbers of this species. However, with a similar distribution and habitat to the Mt Donna Buang wingless stonefly *Riekoperla darlingtoni*, restricted to small upland streams on Mt Donna Buang, this species should be similarly classified as Vulnerable under IUCN (1994) Criterion D2 with a population

> *characterised by an acute restriction in its area of occupancy (typically less than 100 km^2) or in the number of locations (typically less than 5). Such a taxon would thus be prone to the effects of human activities (or stochastic events whose impact is increased by human activities) within a short period of time in an unforeseeable future, and is thus capable of becoming Critically Endangered or even extinct in a very short period.* (IUCN 1994: 20)

The taxon would thus be suitable for listing under the Flora and Fauna Guarantee Act 1988.

Spathula agelaea

This species appears to be present at widespread localities on Mt Buffalo in the National Park and Mt Hotham–Mt Bogong area in ski resort areas, National Park and private grazing areas. It appears to be as widespread as *S. tryssa* and deserves the same IUCN conservation status (Lower Risk Least Concern).

Reynoldsonia reynoldsoni

Although only located on Mt Buffalo, this species is widespread over the plateau and should be considered as secure (Lower Risk Least Concern).

Species 1

Species 1 is the species with the most restricted distribution found in this study. Only found at two sites (probably representing a single connected population), it is clearly under threat from extinction, particularly if horse riding, grazing or other disturbance affects the known habitats. This species should be classified as Critically Endangered under the IUCN categories, and would be suitable for listing under the *Flora and Fauna Guarantee Act* 1988.

Species 2

The distribution of Species 2, while found in a relatively small area around Mt Hotham, cannot be determined as it was spread over the entire sampling area. It is possible that the range extends some considerable distance further east or north. Without adequate data on the range of the species, it should be considered as Data Deficient, defined by the IUCN as a species where 'there is inadequate information to make a direct, or indirect, assessment of its risk of extinction based on its distribution and/or population status' (IUCN 1994: 14). As the species is present at a number of sites within the Alpine National Park, the impact of grazing is likely to be similar to that on *S. tryssa*.

Species 3

Species 3 was found in two well separated diserete areas, so may be seen as widespread. It appears to be as widespread as *S. tryssa* and deserves the same IUCN conservation status (Lower Risk Least Concern).

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